DRUG DISCOVERY

The effect of the imidazoline receptor agonist rilmenidine on visceral and thermal nociceptive pain in mice

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To Cite:
Abdel-Salam OME, Sleem AA. The effect of the imidazoline receptor agonist rilmenidine on visceral and thermal nociceptive pain in mice. *Drug Discovery* 2023; 17: e1dd1001
doi: https://doi.org/10.54905/disssi.v17i39.e1dd1001

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Peer-Review History
Received: 19 October 2022
Reviewed & Revised: 25/October/2022 to 02/January/2023
Accepted: 05 January 2023
Published: 12 January 2023

Peer-Review Model
External peer-review was done through double-blind method.

Drug Discovery
pISSN 2278–540X; eISSN 2278–5396

URL: https://www.discoveryjournals.org/drugdiscovery

ABSTRACT

The effect of the centrally acting imidazoline agonist rilmenidine was examined in acute visceral and thermal nociceptive pain models, Porsolt's forced swimming test and rotarod test in mice. Rilmenidine given intraperitoneally (i.p.) at doses of 0.25, 0.5, 1, 2 or 4 mg/kg dose-dependently inhibited the development of abdominal constrictions evoked by i.p. injection of dilute acetic acid, reducing the number of writhes by 38.4–99.8%. The antinociceptive effect of rilmenidine (1 mg/kg, i.p.) was reduced by atropine, the non-selective beta-adrenoceptor blocker propranolol, the α-2 adrenoceptor antagonist yohimbine and the ATP-sensitive potassium channel blocker glibenclamide. The antinociception caused by rilmenidine was enhanced by the α1-adrenoceptor antagonist prazosin and the sympathetic blocker guanithidine. The drug at the dose of 1 or 2 mg/kg produced significant increases in response latencies in the hot plate test. Rilmenidine (0.5-2 mg/kg, i.p.) did not alter the immobility time in the Porsolt's forced-swimming test or time spent on rotarod testing. The number of spontaneous movements was not significantly affected by rilmenidine at 0.5, 1 or 2 mg/kg, i.p. These results indicate that rilmenidine exerts antinociceptive action in thermal and visceral inflammatory pain models in mice. The visceral pain inhibitory action of rilmenidine may involve ß-adrenergic, cholinergic and K_{ATP} channels.

Keywords: Rilmenidine, imidazoline receptors, visceral pain, thermal pain, hot-plate, writhing

1. INTRODUCTION

Rilmenidine is a centrally acting antihypertensive drug, a structural analogue of clonidine having an oxazolime group which is very similar to the imidazoline moiety. It acts on imidazoline type I receptors in rostral ventrolateral medulla and to much less degree binds to α_2-adrenergic receptors. Imidazoline receptors are nonadrenergic binding sites that recognize compounds having an imidazoline moiety (Bousquet and Feldman, 1999) and subclassified as I_1-, I_2- and I_3-sites (Eglen et al., 1998; Bousquet, 1997). The imidazoline receptors are involved in reduction of central sympathetic tone. The ability of rilmenidine to reduce blood pressure is ascribed to both central and peripheral mechanisms of action. The drug by binding to α_2-adrenoceptors and imidazoline receptors in brain decreases

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central sympathetic outflow. The release of catecholamines from the adrenal medulla is also decreased. The overall effect of rilmenidine is therefore the decrease of sympathetic tone (Bousquet and Feldman, 1999; Bousquet et al., 2020).

The rostral ventromedial medulla is the main source of descending pathways to the spinal cord that can enhance or inhibit central nociceptive processing (Zhou and Gebhart, 2002; Vanegas and Schaible, 2004; Marshall et al., 2012). Rilmenidine has been reported to exert antinociceptive effects. Sabetkasaie et al., (2004) showed that the drug at doses of 1.25, 2.5 and 5 mg/kg, i.p. induced analgesia in the formalin test in mice. Rilmenidine also enhanced the analgesia produced by the non-steroidal anti-inflammatory drug (NSAID) ibuprofen in mouse writhing assay (Soukupova et al., 2009). The aim of this study was therefore to examine the effect of rilmenidine on acute thermal and visceral pain in mice and attempt to elucidate further the neural pathways possibly involved in modulation of its analgesic effect.

2. MATERIALS AND METHODS

Animals

Experiments were performed using male Swiss albino mice weighing 23-25 g of body weight. Mice were housed under standardized conditions with free access to food and water. Experimental procedures were performed in accordance with the Ethics Committee of the National Research Centre and the Guide for Care and Use of Laboratory Animals by the U.S. National Institutes of Health (Publication No. 85-23, revised 1996). Experimental groups of six mice each were tested. Experiments were performed between 9:00 and 11:00 h. Mice were used only once in this study.

Drugs

The drugs and chemicals used in the study were: Rilmenidine (Servier, Egypt), guanethidine, propranolol hydrochloride, prazosin, yohimbine hydrochloride, (Sigma, St. Louis, U.S.A.), glibenclamide (Hoechst Orient, Cairo), atropine sulphate (Misr Pharm Co., Cairo), Analytical-grade glacial acetic acid (Sigma, St. Louis, U.S.A.) is diluted with pyrogen-free physiological saline solution to provide a 0.6% solution for the i.p. injection.

Acetic acid-induced writhing assay

Rilmenidine (0.25, 0.5, 1, 2 or 4 mg/kg) or saline was injected i.p.30 min before an injection of dilute acetic acid (0.6%, 0.2 ml, i.p.). Each mouse was placed in a clear plastic observational chamber. The number of writhes was recorded between 0 and 30 min after the injection of acetic acid (Koster et al., 1959). The observer was not aware of the animals' treatments. Animals were euthanized by CO_2 exposure.

Other experiments were designed to elucidate a possible role for adrenergic, cholinergic or K^+- channels mechanisms in rilmenidine antinociception. For the antagonists experiments, the α-1 adrenoceptor antagonist prazosin (1 mg/kg, i.p.), the α-2 adrenoceptor antagonist yohimbine (4 or 8 mg/kg, i.p.), the non-selective β-adrenoceptor antagonist propranolol (1 or 2 mg/kg, i.p.), the sympathetic neuron blocker guanithidine (8 or 16 mg/kg i.p.), the muscarinic acetylcholine receptor antagonist atropine (1 or 2 mg/kg i.p.), or the K^+- channel blocker glibenclamide (5 or 10 mg/kg i.p.) were co-injected i.p. with rilmenidine (1 mg/kg, i.p.). Acetic acid injections were carried out 30 min after the injection of drugs.

Hot-plate test

The test was performed with the use of an electronically controlled hotplate apparatus (Ugo Basile, Italy) heated to 53°C (± 0.1°C). Baseline measurements were recorded just prior to administering saline or rilmenidine. Mice were treated with i.p. saline or rilmenidine (0.125, 0.25, 0.5 or 1 mg/kg) and 1 h later, the latency to lick the hind paw or jump out of the apparatus was recorded for both the saline and rilmenidine-treated groups. The cut-off time was 30s (Le Bars et al., 2001).

Locomotor activity test

Spontaneous locomotor activity of the mice was measured by an activity monitor equipped with photoelectric detectors (Ugo Basile, Italy). Groups of mice were administered either saline or different doses of rilmenidine (0.5, 1 or 2 mg/kg, i.p.) 30 min before testing. Each mouse was individually placed into the plastic cage and the number of spontaneous locomotor activity (the total number of horizontal photobeam breaks) was counted over a 6-min time period for each animal.

Rotarod testing

Mice motor performance was assessed using rotarod located over plates connected to an automatic counter (Ugo Basile, Varese, Italy). The latency to fall from an accelerating rotarod was determined for the vehicle- and rilmenidine (0.5, 1 or 2 mg/kg, i.p.) treated groups. The time was measured from the beginning of the acceleration period (Millan et al., 1994).

Porsolt's forced-swimming test

Each mouse was forced to swim for 6 min in a glass cylinder that measured 12 cm in diameter and 24 cm height and filled with water maintained at 25°C. Time the mouse spent floating and immobile was considered a measure of despair (Porsolt et al., 1977). Mice were treated with rilmenidine (0.5, 1 or 2 mg/kg, i.p) or saline, 30 min before the procedure.

Statistical analysis

One-way ANOVA followed by Tukey's multiple comparisons test was used for analysis of the data. Graph Pad Prism 6 for Windows (Graph Pad Prism Software Inc., San Diego, CA, USA) was used. Differences between means were considered statistically significant if probability value was < 0.05.

3. RESULTS

Hot-plate test

The reaction time on the hot-plate was significantly delayed after the administration of rilmenidine at 1 or 2 mg/kg, compared with the respective basal values, denoting decreased thermal nociception (Figure 1).

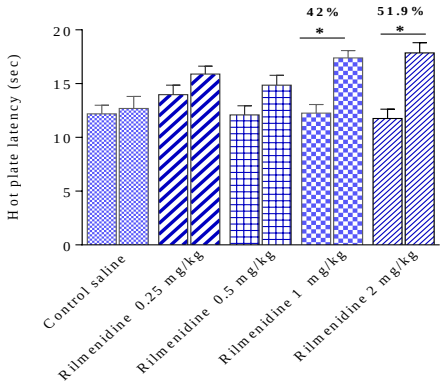

Figure 1 Effect of rilmenidine in hot-plate test in mice. Data represent mean ± SE and percent of increase in hot plate latency compared with the respective basal value. *p < 0.05 *vs.* respective basal value

Writhing test

The number of abdominal constrictions evoked by i.p. acetic acid injection were significantly inhibited in mice pretreated with i.p. rilmenidine at doses of 0.25, 0.5, 1, 2 or 4 mg/kg. This antinociceptive effect of rilmenidine was dose-related with a maximal reduction of the writhing score by 99.8% after rilmenidine administration at 4 mg/kg (Figure 2). The dose of 1 mg/kg of rilmenidine was selected to be used in the subsequent antagonist experiments.

The antinociceptive action of rilmenidine (1 mg/kg, i.p.) was significantly reduced by the co-administration of atropine, propranolol, or yohimbine. Glibenclamide given at 5 mg/kg antagonized the rilmenidine anti nociception while the higher dose of 10 mg/kg enhanced the rilmenidine effect (Figure 3). On the other hand, rilmenidine-induced anti nociception was augmented by the co administration of guanithidine or prazosin (Figure 4).

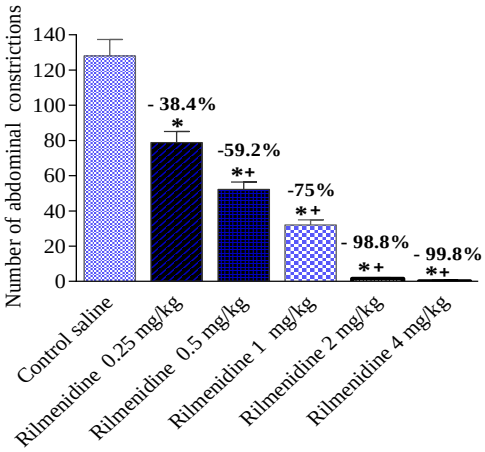

Figure 2 Effect of rilmenidine in the mouse writhing assay induced by i.p. acetic acid. Data represent mean ± SE and percent of inhibition of number of abdominal constrictions compared with saline control. *p < 0.05 *vs.* rilmenidine control, +p <0.05 vs. rilmenidine 0.25 mg/kg treated group

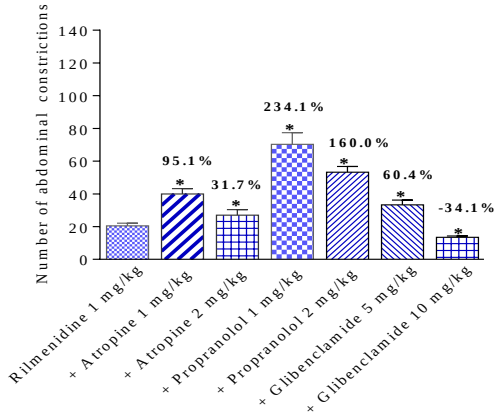

Figure 3 Effect of atropine, propranolol and gibenclamide on antinociception induced by i.p. rilmenidine in the mouse writhing assay. Data represent mean ± SE and percent of inhibition of number of abdominal constrictions compared with rilmenidine control. *p < 0.05 *vs.* rilmenidine control

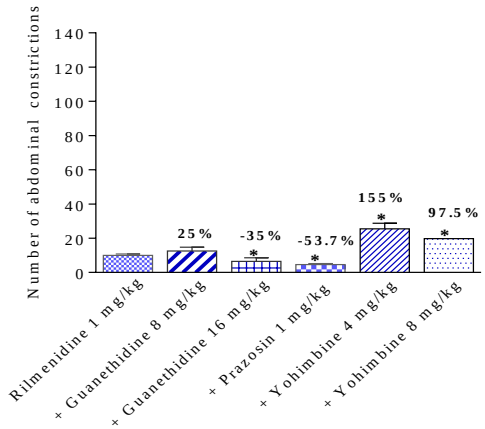

Figure 4 Effect of guanithidine, prozosin and yohimbine on antinociception induced by i.p. rilmenidine in the mouse writhing assay. Data represent mean ± SE and percent of inhibition of number of abdominal constrictions compared with rilmenidine control. *p < 0.05 *vs.* rilmenidine control

Spontaneous locomotor activity

Compared with the saline control, the number of spontaneous movements was not significantly altered by rilmenidine given at 0.5 or 1 mg/kg. The number of spontaneous movements decreased by -20.2% after treatment with 2 mg/kg rilmenidine but this effect did not reach statistical significance (Figure 5A).

Rotarod testing

Rilmenidine at doses of 0.5, 1 or 2 mg/kg did not produce any significant effect on the rotarod performances of mice. No significant differences were observed in the latency to fall between mice treated with rilmenidine and their controls (Figure 5B).

Porsolt's forced-swimming test

As shown in figure 5, treatment with rilmenidine at doses of 0.25-2 mg/kg didn't significantly alter the immobility time (Figure 5C).

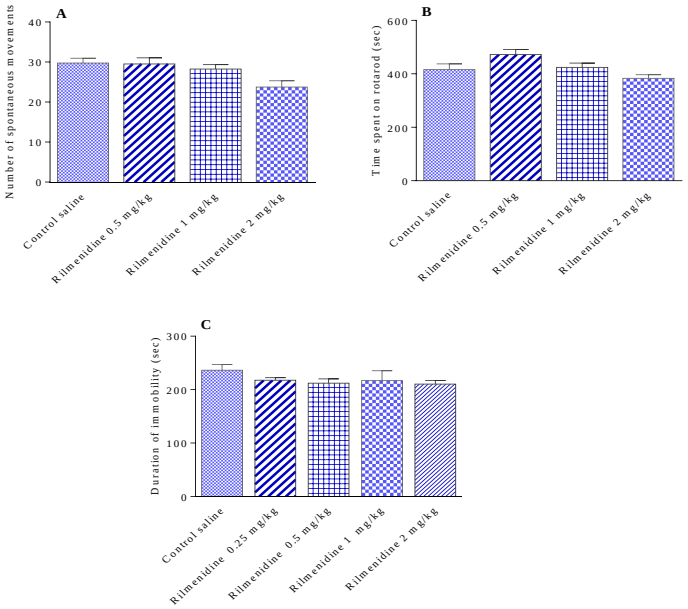

Figure 5 Effect of rilmenidine on (A) spontaneous activity mice; (B) the time spent by mice on the accelerating rotarod; (C) the duration of immobility in Porsolt's forced-swimming test.

4. DISCUSSION

The present experiments showed that the systemic administration of the centrally acting imidazoline receptor agonist rilmenidine produced analgesia in both acute visceral and thermal pain models in mice. Rilmenidine had no significant effect on spontaneous motor activity or the ability of mice to keep their balance in the rota rod test, thereby, indicating that a sedative effect does not contribute to the observed antinociception. The lack of a significant sedative effect for rilmenidine at doses up to at doses up to 10 mg/kg has been reported in mice and rats and attributed to the high affinity for medullary imidazoline binding sites than for α_2-adrenoceptors (Bricca et al., 1989; Montastruc et al., 1989).

Noxious heat-induced pain is mediated by primary afferent nociceptors located on the nerve terminals of sensory neurons with unmyelinated C and thinly myelinated Aδ fiber afferents (Steeds, 2016; Hladnik et al., 2015). In the present study, we found that the paw withdrawal latencies in response to heat increased by rilmenidine. Visceral nociceptive afferents that innervate the gut are also Aδ and C fibres and accompany sympathetic nerves to enter the spinal cord at the dorsal horn nociceptive fibers from the viscera accompany sympathetic nerves to enter the spinal cord at the dorsal horn. Most forms of pain arising from the gastrointestinal tract are mediated by activity in visceral afferent fibres running in sympathetic nerves (Cervero, 1988; Gebhart and Bielefeldt, 2016). Most of the nociceptors found in the viscera, however, are "silent" being activated by inflammatory mediators (Hladnik et al., 2015). The primary afferent nerve fibres have their cell bodies in the dorsal root ganglia and/or cranial sensory nuclei and terminate in the spinal cord dorsal horn with the nociceptive information being ultimately conveyed to the cerebral cortex (Westlund, 2000; Sikanda and Dickenson, 2012).

In this study, we found that the behavioral changes (i.e., writhing) that followed noxious chemical stimulation of viscera by i.p. acetic acid were markedly inhibited by rilmenidine in a dose-dependent fashion. Studies have shown that the writhing response is brought about by the release of prostacyclin into the abdominal cavity (Berkenkopf and Weichman, 1988). Moreover, in response to local tissue injury, there will be a release of inflammatory mediators like bradykinin, prostaglandins and adenosine triphosphate from inflammatory cells and these agents activate spinal, vagal or pelvic afferents that mediate nociceptive signals to the central nervous system (Bueno and Fioramonti, 2002). These nociceptive signals and/or the causative inflammatory process are subject to both descending facilitating and inhibitory modulation from higher centers and limbic structures (Sikanda and Dickenson, 2012). In this context, the writhing response has been shown to be subject to inhibition by a variety of drugs which inhibits the synthesis of prostaglandins eg., the non-steroidal anti-inflammatory drug (NSAID) meloxicam (Santos et al., 1998), drugs that modulates brain neurotransmitters eg., fluoxetine (Singh et al., 2001), cinnarizine (Abdel-Salam, 2007a), opioid receptor agonists eg., morphine (Baamonde et al., 1989), the transient receptor potential cation channel vanilloid subfamily member 1 (TRPV1) agonists such as capsaicin (Abdel-Salam, 2006a) and piperine (Abdel-Salam et al., 2007b), cannabinoids eg., Δ^9-tetrahydrocannabinol and cannabinol (Booker et al., 2009) and antioxidants like melatonin (Abdel-Salam et al., 2006c).

Our results showed that sympathetic neuron blockade with guanithidine and the α1-adrenoceptor antagonist prazosin both augmented the visceral antinociceptive action of rilmenidine. Guanithidine is an adrenergic neuron blocking drug which is taken up by noradrenaline transporters in adrenergic nerve endings and prevents the release of noradrenaline from storage vesicles, thereby, decreasing the sympathetic tone (Ritter, 2018). Guanithidine is an effective agent in the management of sympathetically maintained neuropathic and visceral pain (Neil et al., 1991; Menon and Swanepoel, 2010; Gil et al., 2016). Significant inhibition of the writhing response to i.p. acetic acid in mice was reported after guanethidine (27% inhibition at 30 mg/kg, s.c.) (Duarte et al., 1988). Other studies showed that guanithidine given at 8 or 32 mg/kg by itself inhibited the writhing response by 16.1% and 75.4%, respectively (Abdel-Salam et al., 2006c; Abdel-Salam and Baiuomy, 2008).

The administration of α2- adrenergic receptor agonists produces antinociception by Inhibition of synaptic transmission in the spinal cord dorsal horn (Sabetkasaiea et al., 2004; Tanabe et al., 2005). Our results showed that rilmenidine action was potentiated by prazosin but antagonized by the α-2 adrenoceptor antagonist yohimbine. Sierralta et al., (1996) have shown in the writhing test of mice that prazosin (1-10 mg/kg) exerted antinociceptive action and enhanced the analgesic effect of i.p. clonidine and α1-adrenoceptor agonist. In addition, i.p. yohimbine (0.3-10 mg/kg) was able to antagonize the clonidine antinociception. It is to be noted that clonidine is also able to bind to nonadrenergic imidazoline receptors. Moreover, the action of clonidine and the related drug rilmenidine in reducing central sympathetic tone is thought to involve inhibition of the sympathoexcitatory reticulospinal neurons of the rostroventrolateral medulla (Khan et al., 1999). It is thus possible that the antinoceptive action of clonidine and also rilmenidine involves imidazoline receptors.

The β-adrenoreceptor antagonists were shown to exert antinociceptive effect in the writhing test of rodents (Duarte et al., 1988; Korzeniewska-Rybicka and Plaznik, 2001). When given at a dose of 2 mg/kg (i.p.) propranolol inhibited the writhing response in mice by 41.6% (Abdel-Salam, 2006b). In the present study, propranolol antagonized the rilmenidine–induced visceral antinoception, suggesting that ß-adrenergic mechanisms may be involved. The spinal cholinergic system and muscarinic receptors are involved in pain modulation (Ghelardini et al., 1990). We therefore investigated the effect of the non-selective muscarinic antagonist atropine. Previous studies have shown that atropine produced analgesic effects in tests of nociception in rodents. These effects, however, were observed with very low doses in the range of 1-l00 μg/kg. In contrast, high doses of 5 mg/kg atropine produced hyperalgesia (Ghelardini et al., 1990). In the writhing test in mice, atropine 2, 3 or 6 mg/kg, i.p. was found to increase the number of abdominal constrictions caused by acetic acid in a dose-dependent manner (Abdel-Salam, 2006c; Salam and Baiuomy, 2008). The drug (1 mg/kg, i.p.) also was reported to antagonize antinociception induced by NSAIDs in tail-flick test, a model of acute thermal pain in mice (2003).

Our present results showed that atropine antagonized the antinociception induced by rilmenidine in the writhing assay, indicating that central cholinergic processes may contribute to the rilmenidine action. We also investigated the possible role of adenosine triphosphate-sensitive (K_{ATP}) channels with the use of glibenclamide (10 mg/kg, i.p.), a K_{ATP} channel blocker. These channels have been implicated in the modulation of visceral pain processing (Han et al., 2004; Qian et al., 2009). Blockade of with gibenclamide (5 mg/kg, i.p.) was found to inhibit abdominal writhing in mice (Abdel-Salam 2006b; 2007a). In the present study, the administration of glibenclamide at 5 mg/kg was found to antagonize the action of rilmenidine. The higher dose, however, enhanced antinociception by rilmenidine. These data suggest that K_{ATP} channels may be also involved in the mechanisms by which rilmenidine exerts its visceral antinociceptive action.

5. CONCLUSION

The present study provided the first evidence that the imidazoline receptor agonist rilmenidine exerts antinociceptive effects against thermal and visceral inflammatory (chemogenic) pain in mice. The mechanism of the visceral antinociceptive activity of rilmenidine may involve ß-adrenergic, cholinergic and K_{ATP} channels.

Author contribution: O.M.E.A.S. and A.A.S. conducted the research. O.M.E.A.S. wrote and prepared the manuscript, O.M.E.A.S. and A.A.S. approved the final version of the manuscript.

Ethical approval

Experimental procedures were performed in accordance with the Ethics Committee of the National Research Centre and the Guide for Care and Use of Laboratory Animals by the U.S. National Institutes of Health (Publication No. 85-23, revised 1996).

Informed consent: Not applicable.

Conflicts of interests

The authors declare that there are no conflicts of interests.

Funding

The study has not received any external funding.

Data and materials availability

All data associated with this study are present in the paper.

REFERENCES AND NOTES

1. Abdel Salam OME, EL-Shenawy S, Nofal SM, Mózsik Gy. Piperine inhibitis visceral pain caused by acetic acid in mice. J Pharmacol Toxicol 2007b; 2:456-464.
2. Abdel Salam OME. Vinpocetine and piracetam exert antinociceptive effect in visceral pain model in mice. Pharmacol Rep 2006b; 58:680-691.
3. Abdel-Salam OME, Baiuomy AR. Citric acid strongly inhibits visceral pain response in mice. Excli J 2008; 7:93-103.
4. Abdel-Salam OME, El-Batran SA, Baiuomy AR. Noradrenergic and dopaminergic modulation of melatonin visceral antinociception in mice. J Pharmacol Toxicol 2006c; 1: 234-244.
5. Abdel-Salam OME. Modulation of visceral nociception by capsaicin in mice. J Pharmacol Toxicol 2006a; 1:493-504.
6. Abdel-Salam OME. Modulation of visceral nociception, inflammation and gastric mucosal injury by cinnarizine. Drug Target Insights 2007a; 2:29–38.
7. Baamonde A, Hidalgo A, Andres-Trelles F. Sex-related differences in the effects of morphine and stress on visceral pain. Neuropharmacology 1989; 28:967–970.
8. Berkenkopf JW, Weichman BM. Production of prostacyclin in mice following intra peritoneal injection of acetic acid, phenyl benzoquinone and zymosan: Its role in the writhing response. Prostaglandins 1988; 36:693–709.
9. Booker L, Naidu PS, Razdan RK, Mahadevan A, Lichtman AH. Evaluation of prevalent phytocannabinoids in the acetic acid model of visceral nociception. Drug Alcohol Depend 2009; 105(1-2):42–47.
10. Bousquet P, Feldman J. Drugs acting on imidazoline receptors. A review of their pharmacology, their use in blood pressure control and their potential interest in cardioprotection. Drugs 1999; 58(5):799-812.
11. Bousquet P, Hudson A, García-Sevilla JA, Li JX. Imidazoline receptor system: The Past, the present and the future. Pharmacol Rev 2020; 72(1):50–79.
12. Bousquet P. I_1-imidazoline receptors: From the pharmacological basis to the therapeutic application. J Hypertens Suppl 1997; 15(1):S9–23.
13. Bricca G, Dontenwill M, Molines A, Feldman J, Tibirica E, Belcourt A, Bousquet P. Rilmenidine selectivity for imidazoline receptors in human brain. Eur J Pharmacol 1989; 163(2-3):373-377.
14. Bueno L, Fioramonti J. Visceral perception: Inflammatory and non-inflammatory mediators. Gut 2002; 51(Suppl I):i19–i23.
15. Cervero F. Neurophysiology of gastrointestinal pain. Baillieres Clin Gastroenterol 1988; 2(1):183–99.
16. Duarte ID, Nakamura M, Ferreira SH. Participation of the sympathetic system in acetic acid-induced writhing in mice. Braz J Med Biol Res 1988; 21:341–343.
17. Eglen RM, Hudson AL, Kendall DA, Nutt DJ, Morgan NG, Wilson VG, Dillon MP. Seeing through a glass darkly: Casting

light on imidazoline "I" sites. Trends Pharmacol Sci 1998; 19:381–390.
18. Gebhart GF, Bielefeldt K. Physiology of visceral pain. Compr Physiol 2016; 6:1609–1633.
19. Ghelardini C, Malmberg-Aiello P, Giotti A, Malcangio M, Bartolini A. Investigation into atropine-induced antinociception. Br J Pharmacol 1990; 101:49–54.
20. Gil DW, Wang J, Gu C, Donello JE, Cabrera S, Al-Chae ED. Role of sympathetic nervous system in rat model of chronic visceral pain. Neurogastroenterol Motil 2016; 28(3):423–431.
21. Han BF, Zhang C, Reyes-Vazquez C, Qiao JT, Dafny N. ATP-sensitive potassium channels and endogenous adenosine are involved in spinal antinociception produced by locus coeruleus stimulation. Int J Neurosci 2004; 114:961–974.
22. Hladnik A, bičanić I, Petanjek Z. Functional neuroanatomy of nociception and pain. Period biol 2015; 117(2):195–204.
23. Khan ZP, Ferguson CN, Jones RM. alpha-2 and imidazoline receptor agonists. Their pharmacology and therapeutic role. Anaesthesia 1999; 54(2):146–165.
24. Korzeniewska-Rybicka I, Plaznik A. Role of serotonergic and noradrenergic systems in a model of visceral pain. Pol J Pharmacol 2001; 53:475–480.
25. Koster R, Anderson M, De Beer EJ. Acetic acid for analgesic screening. Fed Proc 1959; 18:412.
26. Le Bars D, Gozariu M, Cadden SM. Animal models of nociception. Pharmacol Rev 2001; 53:597–652.
27. Marshall TM, Hermana DS, Largent-Milnes TM, Badghisi H, Zuber K Holt SC, Lai J, Porreca F, Vanderah TW. Activation of descending pain facilitatory pathways from the rostral ventromedial medulla by cholecystokinin elicits release of PGE_2 in the spinal cord. Pain 2012; 153(1):86–94. doi: 10.1016/j.pain.2011.09.021.
28. Menon R, Swanepoel A. Sympathetic blocks, Continuing Education in Anaesthesia Critical Care & Pain 2010; 10(3):88-92. doi: 10.1093/bjaceaccp/mkq012
29. Millan MJ, Bervoets K, Rivet JM, Widdowson P, Renouard A, Le Marouille-Girardon S, Gobert A. Multiple α_2 -adrenergic receptor subtypes II. Evidence for a role of rat $R_2\alpha$ -ARs in the control of nociception, motor behavior and hippocampal synthesis of noradrenaline. J Pharmacol Exp Ther 1994; 270:958–972.
30. Montastruc JL, Macquin-Mavier I, Tran MA, Damase-Michel C, Koenig-Berard E, Valet P. Recent advances in pharmacology of rilmenidine. Am J Med 1989; 87(suppl 3C):3c-14s.
31. Neil A, Attal N, Guilbaud G. Effects of guanethidine on sensitization to natural stimuli and self-mutilating behaviour in rats with a peripheral neuropathy. Brain Res 1991; 565:237-246.
32. Pinardi G, Sierralta F, Miranda HF. Atropine reverses the antinociception of nonsteroidal anti-inflammatory drugs in the tail-flick test of mice. Pharmacol Biochem Behav 2003; 74 (3):603-608.
33. Porsolt RD, Le Pichon M, Jalfre M. Depression: A new animal model sensitive to antidepressant treatments. Nature 1977; 266:730–732.
34. Qian AH, Liu XQ, Yao WY, Wang HY, Sun J, Zhou L, Yuan YZ. Voltage-gated potassium channels in IB4-positive colonic sensory neurons mediate visceral hypersensitivity in the rat. Am J Gastroenterol 2009; 104:2014-2027.
35. Ritter JM. Noradrenergic transmission. In: Rang & Dale's Pharmacology, 9th Edition, Elsevier 2018; 197-216.
36. Sabetkasaie M, Vala S, Khansefid N, Hosseini AR, Sadat Ladgevardi MA. Clonidine and guanfacine-induced antinociception in visceral pain: possible role of alpha 2/I2 binding sites. Eur J Pharmacol. 2004; 501(1-3):95-101. doi: 10.1016/j.ejphar.2004.08.010
37. Santos AR, Vedana EM, De-Freitas GA. Antinociceptive effect of meloxicam, in neurogenic and inflammatory nociceptive models in mice. Inflamm Res 1998; 47:302–307.
38. Sierralta F, Naquira D, Pinardi G, Miranda HF. α-Adrenoceptor and opioid receptor modulation of clonidine induced antinociception. Br J Pharmacol 1996; 119:551–554.
39. Sikandar S, Dickenson AH. Visceral pain – the ins and outs, the ups and downs. Curr Opin Support Palliat Care 2012; 6 (1):17–26. doi: 10.1097/SPC.0b013e32834f6ec9
40. Singh VP, Jain NK, Kulkarni SK. On the antinociceptive effect of fluoxetine, a selective serotonin reuptake inhibitor. Brain Res 2001; 915:218–226.
41. Soukupova M, Dolezal T, Krsiak M. Synergistic interaction between rilmenidine and ibuprofen in the writhing test in mice. Neuro Endocrinol Lett 2009; 30(2):215-20.
42. Steeds CE. The anatomy and physiology of pain. Surgery 2016; 34(2):55-59.
43. Tanabe M, Takasu K, Kasuya N. Role of descending noradrenergic system and spinal alpha2-adrenergic receptors in the effects of gabapentin on thermal and mechanical nociception after partial nerve injury in the mouse. Br J Pharmacol 2005; 144:703–714.
44. Vanegas H, Schaible H-G. Descending control of persistent pain: Inhibitory or facilitatory? Brain Res Rev 2004; 46:295–309.
45. Westlund KN. Visceral nociception. Curr Rev Pain 2000; 4(6): 478–487. doi: 10.1007/s11916-000-0072-9
46. Zhou M, Gebhart GF. Facilitation and attenuation of a visceral nociceptive reflex from the rostroventral medulla in the rat. Gastroenterology 2002; 122:1007–1019.